

Peach seedling growth with mycorrhiza and vermicompost

Crecimiento de plántulas de durazno con micorrizas y vermicomposta

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Resumen

La esterilización de sustratos para vivero disminuye la población de microorganismos benéficos en el medio que rodea la raíz y puede resultar en un pobre crecimiento de la planta; la vermicomposta y los hongos micorrízicos arbusculares (HMA) podrían mejorar su desarrollo. El objetivo de este experimento fue analizar el efecto de la vermicomposta y la inoculación de HMA sobre el crecimiento inicial de plántulas de durazno [*Prunus persica* (L.) Batsch.] en sustrato esterilizado. Plántulas de durazno germinadas en perlita estéril fueron distribuidas en cuatro tratamientos resultado de la combinación de dos factores y dos niveles cada uno: con/sin vermicomposta en el sustrato y con/sin inoculación con HMA al momento del trasplante. Las plántulas se distribuyeron completamente al azar dentro de un invernadero de vidrio durante el estudio. La incorporación de vermicomposta al sustrato y la inoculación de HMA, así como su combinación, resultaron en un decremento de clorofila total ($p < 0.05$) a los 108 días después de plantación (DDP). Al final del experimento (180 DDP), la inoculación con HMA resultó en una colonización de raíces mayor al 70% de su longitud total, pero este efecto fue eclipsado por la vermicomposta. El peso seco de tallo y raíz y el diámetro de tallo fueron superiores ($p < 0.01$) con el uso de vermicomposta, pero la inoculación de HMA no tuvo efecto en estas variables. Se concluye que es más recomendable la vermicomposta que la inoculación con HMA para estimular el crecimiento de plántulas de durazno durante los primeros seis meses en sustrato esterilizado.

Palabras clave: *Prunus persica* (L.) Batsch, *Glomus* spp, fotosíntesis, fertilizante orgánico.

Abstract

Sterilization of nursery substrate materials decreases the beneficial microorganisms in the surrounding root media and may result in poor seedling growth; compost and arbuscular mycorrhizal fungi (AMF) may improve the plantlet development. This experiment aimed to analyze the effect of both AMF inoculation and vermicompost for the initial peach [*Prunus persica* (L.) Batsch.] seedling growth in sterilized substrate. Peach seedlings germinated in sterilized perlite were distributed into four resulting treatments from the combination of two factors and two levels each: with/without vermicompost in the growing media and with/without AMF inoculation at the transplanting time. Seedlings were arranged completely randomized inside a glasshouse throughout the study. Utilization of vermicompost in the growing media and AMF inoculation, and their combination, resulted in less total chlorophyll ($p < 0.05$) measured at 108 days after planting (DAP). At the end of the experiment (180 DAP), AMF inoculation resulted in root colonization greater than 70% of the total root length; however, this effect was eclipsed by adding vermicompost to the substrate. Root and shoot dry weights and also stem diameter were superior ($p < 0.01$) by adding vermicompost to the growing substrate, but AMF inoculation had no effect on these variables. It is concluded that vermicompost addition to the substrate is preferable to AMF inoculation in order to stimulate peach seedling growth during the initial six months in sterilized substrates.

Keywords: *Prunus persica* (L.) Batsch, *Glomus* spp, photosynthesis, organic fertilizer.

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Introduction

Sterilization of growing substrates is a necessary activity in seedling greenhouse production to decrease soil borne diseases; however, this activity also could negatively impact plant growth and development in the nursery. Chemical fumigation by chloropicrin and methyl bromide, which is still used in some countries, causes stunting and poor growth in peach seedlings [*Prunus persica* (L.) Batsch.], which may be related to mineral deficiencies in plants (La Rue *et al.*, 1975; Lambert *et al.*, 1979).

Similarly, soils collected from peach nurseries and either autoclaved at 105 °C (40 min), formaldehyde-treated or exposed to ozone, yield stunted peach seedlings in pots (Bingye and Shengrui, 1998). In some cases, further analysis of treated substrates indicated that elimination of beneficial microorganisms, including mycorrhizal fungi, was the primary factor in sterilized soils to cause stunting and growth problems in peach seedlings (Lambert *et al.*, 1979).

After sterilization, soil replenishment with beneficial microorganisms like arbuscular mycorrhizal fungi (AMF) can be achieved by artificially inoculating the substrate. La Rue *et al.* (1975) found that after sterilization with methyl bromide, only mycorrhizal inoculation of the substrate can result in peach colonized roots. Similarly, micropropagated plantlets of peach grown in a sterilized peat-sand mix were able to bear colonized roots only when inoculated with the *Glomus* spp AMF during the early stage of acclimation, while non-inoculated plantlets showed no colonization (Rapparini *et al.*, 1994). In the lilaceous *Brodiaea laxa* 'Queen Fabiola', mycorrhizal inoculation following substrate pasteurization can increase root colonization by between 20% and 45% compared to the non-inoculated and pasteurized treatments (Scagel, 2004). In all these cases plant growth was significantly improved as a result of the fungal root colonization.

The effect of mycorrhizal inoculation in nursery substrates on plant growth and development has been studied in a broad sense. AMF inoculation to the substrate results in significantly higher levels of foliar zinc in peach

seedlings compared to non-inoculated plants (La Rue, 1975); however, this effect could not be observed by inoculating apple (*Malus pumila* Mill.) with AMF and no differences in any mineral foliar content could be observed between inoculated and non-inoculated plants three months after transplanting to the field (Plenchette *et al.*, 1981). In another experiment with *Brodiaea laxa* 'Queen Fabiola' grown in pasteurized substrates during the entire first growing cycle, AMF inoculation resulted in higher corm concentrations of nitrogen (N), potassium (K) and zinc (Zn), but not phosphorus (P) or sulfur (S) (Scagel, 2004). Mycorrhizal influence in plant growth has also been documented and the information includes significant increases in shoot height (La Rue *et al.*, 1975; Plenchette *et al.*, 1981; and Rapparini *et al.*, 1994), root volume (Plenchette *et al.*, 1981), stem diameter (La Rue *et al.*, 1975; Plenchette *et al.*, 1981) and leaf surface area and dry mass (Plenchette *et al.*, 1981).

Regarding compost or vermicompost, its interest as a nursery substrate is more recent than that of mycorrhizae; nevertheless, utilization of these materials as an option for sustainable and safer horticultural crops has increased rapidly in the last few decades. Most growing media is based on peat and mineral products; however, utilization of peat has led to discussions on the deleterious environmental effect that peat extraction can cause as peat bogs regenerate too slowly to support its extraction (Rivière *et al.*, 2008). Because of this concern, investigations are being conducted to find peat substitutes as suitable growing media, and compost or vermicompost are considered

one of the major candidates to replace peat in growing media mixtures (Lanzi *et al.*, 2009). Compost can increase cation exchange capacity (CEC), pH, electrical conductivity (EC), and mineral availability (N, P, K, Ca and Mg) in substrates, thus offering better characteristics to growing media than substrates based solely on peat, perlite and vermiculite, even though it may lower water-holding capacity (Moore, 2004). Vermicomposting, a process whereby organic residues are further broken down by earthworms, can improve the compost characteristics by making them more stable, reducing EC (Lazcano *et al.*, 2008) and increasing total N, available P, exchangeable K, Ca and Mg contents (Suthar and Singh, 2008).

Mycorrhizal inoculation is being used along with compost or vermicompost in order to improve plant mineral uptake and growth, and diverse effects have been found. AMF like *Glomus intraradices* can interact synergistically with high humic content composts and enhance onion plant growth (Linderman and Davis, 2001); however, in peach x almond hybrid rootstocks there is no synergistic effect for the mixture AMF-compost; in this case, adding compost to the growing media can even eclipse the enhancing effect of mycorrhizal inoculation on shoot dry weight and stem diameter (Estaún *et al.*, 1999). Similarly, AMF (*Glomus* spp.) applied along with 5 and 10 t ha⁻¹ of rice straw either composted or vermicomposted, resulted in no synergistic effect on sorghum plant growth (Hameeda *et al.*, 2007). Apparently, the raw material for compost or vermicompost, the composting process, the compost rate, the crop species and the AMF inoculum type and rate could together play an important role in the effect of AMF-compost/vermicompost mixtures on plant development. There is not enough information on the effect of the mixture AMF-vermicompost on peach seedling growth and this information could be important to make decisions about the plant management at nurseries. The objective of this research was to evaluate the effect of vermicompost and the AMF *Glomus* spp. on peach seedling growth on sterilized substrate during the first six months after germination.

Materials and methods

The experiment was performed in a glasshouse in central Mexico (19° 29' 05" LN; 98° 53' 11" LW; 2, 250 m a.s.l.). Temperatures inside the glasshouse fluctuated between 20 and 23 °C during the experiment.

Vermicompost was made with oat straw decomposed during the cultivation of the edible *Pleurotus* fungus. Earthworm *Eisenia foetida* was later added to the compost to stabilize the product. A mixture of the AMF *Glomus* spp. (*Glomus albidum* Walker & Rhodes, *Glomus diaphanum* Schenck & Smith, and *Glomus claroide* Morton & Walker) was used. This particular mixture was isolated in a sandy soil (pH 5.6) previously cultivated with bean in the state of Zacatecas (north-central Mexico). Inoculum consisted of sorghum-colonized roots with 68.6% colonization and 285 *Glomus* spores per 100 g of inoculum.

Peach seeds [*Prunus persica* (L.) Batsch] from a commercial nursery were treated with a solution of clorox 10% and distilled water, then chilled for 500 h at 4 °C in autoclaved sand. After chilling, seeds were moved into a glasshouse and germinated in autoclaved expanded perlite. After germination, 68 seedlings about 7.0 cm tall were transplanted. Half of the seedlings were transplanted into a growing media consisting of organic forest soil, sand and expanded perlite (1:1:1, v/v), while the remaining half of the seedlings were transplanted in a growing media consisting of vermicompost, organic forest soil, sand and expanded perlite (3:1:1:1, v/v). Except for vermicompost, all materials for growing media, seed chilling and germination were autoclaved at 1.4 kg cm⁻² for three hours prior to use. Each growing media (with and without vermicompost) was analyzed for chemical characteristics (Table 1). During transplanting, half of the seedlings from each growing media were inoculated with 10 g plant⁻¹ of the AMF *Glomus* spp. inoculum and the other half remained non-inoculated. Thus, the experiment consisted of four growing media treatments: no vermicompost with no AMF inoculation (control),

no vermicompost with AMF inoculation, vermicompost and no AMF inoculation, and vermicompost with AMF inoculation. Inoculation was done after the seedling was placed in the corresponding growing media by positioning the inoculum around the seedling root. Treatments were arranged inside the glasshouse in a completely randomized design. Experiment began with 17 replications per treatment; however, because of destructive samplings throughout the experiment, replications for treatments varied among sampling days.

Table 1. Chemical characteristics at the beginning of the trial for two tested growing media effects on peach seedling growth.

Factor	Growing Media ²	
	With Vermicompost	Without Vermicompost
pH	6.98	6.01
Electrical Conductivity (dS·m ⁻¹)	7.47	0.89
Organic Matter (%)	5.38	2.82
N (mg·kg ⁻¹)	70.00	42.00
P (mg·kg ⁻¹)	295.00	65.00
K (mg·kg ⁻¹)	1800.00	328.00
Ca(mg·kg ⁻¹)	5076.35	1215.54
Mg (mg·kg ⁻¹)	657.73	156.26
Fe (mg·kg ⁻¹)	27.56	42.37
Cu (mg·kg ⁻¹)	1.01	0.37
Zn (mg·kg ⁻¹)	8.87	1.57
Mn (mg·kg ⁻¹)	59.38	46.01

²Growing media with vermicompost was made of vermicompost, organic forest soil, sand and expanded perlite (3:1:1:1, v/v). Growing media without vermicompost was made of organic forest soil, sand and expanded perlite (1:1:1:1, v/v).

At 94 days after transplant (DAP), plant height, stem diameter and number of leaves per plant were determined on all seedlings of each treatment. At 108 DAP, chlorophyll content, CO₂ assimilation and stomatal conductance were determined from five random plants of each treatment. At the end of the experiment (180 DAP), plant height, stem diameter, root

colonization and leaf N, P and K concentration were determined from seven seedlings of each treatment. At this time, seedlings were separated into roots and shoots, dried to constant weight at 85 °C and dry weights determined. Leaves were sent to laboratory for mineral analysis. Foliar N, P and K were determined by micro-Kjeldahl method (Chapman and Pratt, 1973), wet digestion (Etchevers, 1987) and flame photometry (Model 410 Flame Photometer; Sherwood), respectively.

Collected peach roots at 180 DAP were cleaned from substrate and maintained in FAA (formaldehyde – acetic acid – ethanol). Root colonization was measured based on Phillips and Hayman (1970) procedure. Roots were treated with a solution of KOH 10% and heated in a pressurized oven at 7.03 kg cm⁻² for 10 minutes. Then, roots were rinsed and treated with HCl 10% for 15 min and added with trypan blue 0.05% in lactoglycerol and heated at 7.03 kg cm⁻² for 10 min. Roots were rinsed and observed through a microscope and colonization was estimated based on the number of root segments observed and those which were detected as colonized.

At 108 DAP two discs 3.46 cm² each were obtained from the seventh or eighth fully extended leaf from the apex in each seedling. Discs were placed into a flask with 5 mL acetone 80% and wrapped with aluminum foil until dark centrifugation with an Eppendorf 5415C centrifuge. Chlorophylls a and b were determined with a spectrophotometer Hewlett Packard 8453. In order to determine CO₂ assimilation and stomatal conductance, two fully expanded leaves from each replication were analyzed with a LI-COR LI6200 as a closed system and under full sun light (8:00 am throughout to 10:00 am).

Data was analyzed by the ANOVA procedure with the SAS program (SAS Institute Inc., Cary, NC. USA) under a 2x2 factorial model. In the event of mycorrhiza x vermicompost significant interaction, further analysis was done and the simple effects were detected. Means were separated by the Tukey test.

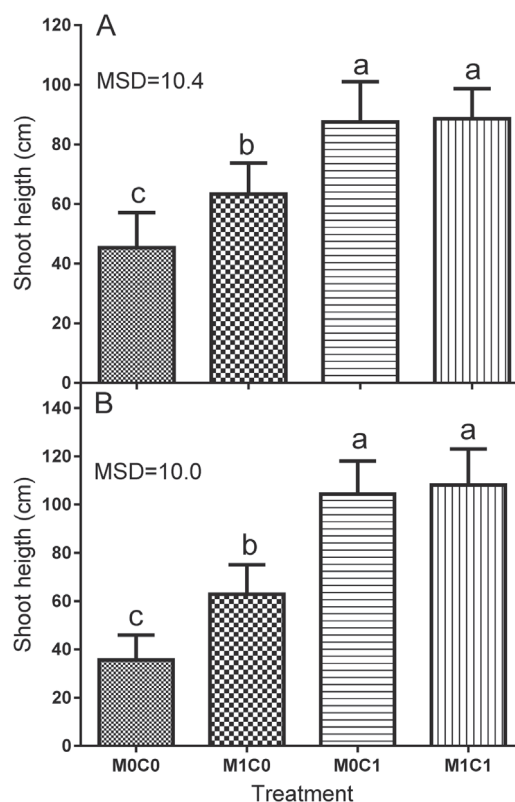
Results and discussion

Mycorrhizal inoculation and vermicompost had no significant interaction effects on half of the variables analyzed in this experiment. In the case of plant height, chlorophyll content, root AMF colonization and leaf N and P concentration, an interaction effect was observed between AMF inoculation and vermicompost ($p < 0.05$, 0.001, 0.0001 and 0.001, respectively). In this case, every combination from the levels of both factors was analyzed and compared to each other in order to make the most appropriate conclusions.

The effect of treatments on plant height observed at 94 DAP was still evident at 180 DAP, thus showing the consistency of the seedling response to AMF inoculation and vermicompost (Figure 1). In this case, the effect of mycorrhizal inoculation depended on the presence of vermicompost in the substrate as AMF inoculation resulted in statistically taller shoots only when plants growing in media without vermicompost are compared. When vermicompost was added to the growing media, shoot height almost doubled that in peach plants growing in media without vermicompost; however, AMF inoculation resulted in no statistical differences for shoot height when plants growing in media with vermicompost are compared.

Mycorrhizal inoculation has been found to enhance peach growth in substrates with low P and K but its effect disappears when plants are grown in compost media, which results in higher amounts of N and P in the substrate (Estaún *et al.*, 1999). Also, mycorrhizal inoculation improved peach seedling growth in autoclaved soils (Bingye and Shengrui, 1998). In this experiment, growing media without vermicompost was autoclaved before transplanting peach seedlings and some improvement of plant growth was expected as a result of AMF inoculation.

Figure 1. Shoot height of peach seedlings as affected by vermicompost addition to the growing substrate and AMF inoculation at transplanting time. A, 94 days after transplanting ($n = 17$); B, 180 days after transplanting ($n = 7$). M0C0, control; M1C0, AMF-inoculated in substrate without vermicompost; M0C1, non-AMF-inoculated in substrate with vermicompost; and M1C1 AMF-inoculated in substrate with vermicompost. Different letters in the date indicate statistical differences by Tukey Test ($\alpha = 0.05$).



When mycorrhizal plants are compared to nonmycorrhizal plants, no differences in stem diameter are detected. This is true for plants at 94 DAP (Figure 2) and also for plants at 180 DAP (Table 2); however, AMF inoculation did result in statistically more leaves per shoot (Figure 3) and statistically less chlorophylls concentration compared with control plants (Figure 4). It appears that more leaves in AMF inoculated plants although with less chlorophylls were just enough to stimulate shoot length but not diameter increases. Rapparini *et al.* (1994) reported significant increases in leaf fresh weight and internode length in AMF inoculated peach plants during the first year of growth compared with non-AMF inoculated plants.

Figure 2. Stem diameter of peach seedlings at 94 days after planting as affected by vermicompost addition to the growing substrate and AMF inoculation at transplanting time. Different letters in the factor indicate statistical differences by Tukey Test ($\alpha = 0.05$; $n = 14$).

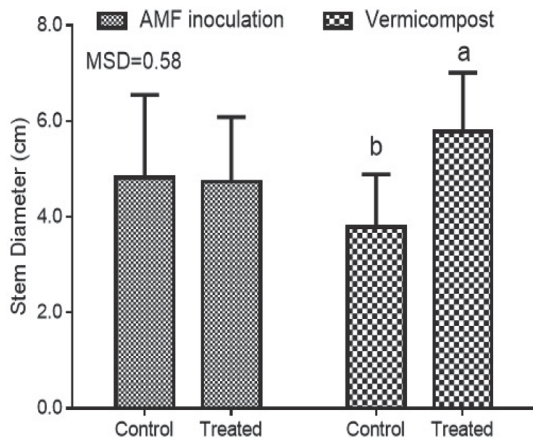


Table 2. Dry weights and stem diameter of peach seedlings as affected by AMF inoculation at transplanting and vermicompost addition to the growing substrate. Data at 180 days after transplanting is the mean ($n = 7$).

Factor	Level	Stem Diameter (cm)	Dry Weight (gr)		
			Shoot	Root	Root/Shoot
AMF inoculation	Control	5.36	12.21	7.07	0.53
	AMF inoculated	5.63	14.81	7.08	0.57
Vermicompost	Control	3.48	2.64	1.85	0.64
	Vermicompost	7.51	26.38	12.30	0.47

Analysis of Variance				
AMF inoculation (M)	ns	ns	ns	ns
Vermicompost (V)	**	**	**	**
MxV	ns	ns	ns	ns

ns, non-significant; and **, high significance ($p < 0.01$) by Tukey test.

Unlike AMF inoculation, vermicompost addition to the substrate resulted in highly significant differences in both stem diameter and leaves per shoot (Table 2; Figures 2 and 3). The main benefits from adding vermicompost to soils are the increase in organic matter, which improves soil biological activity, and soil physical characteristics including soil respiration, enzyme activity, nitrification rate, water infiltration, hydraulic conductivity and water holding capacity (Raviv, 2005) and finally resulting in better plant growth. In this experiment, AMF inoculation did not affect dry weights from shoot, root nor root

to shoot ratio (Table 2), meaning that the increase in leaves per shoot which resulted from AMF inoculation was not sufficient to alter shoot dry weight in these plants. On the other hand, when we compare peach plants grown in media with vermicompost with those grown in media without vermicompost, significant increases in shoot and root dry weights are observed. These increases ranged from 565% to 899% for root and shoot dry weights respectively and resulted in a significant decrease in root to shoot dry weight ratio for plants grown with vermicompost.

Figure 3. Number of leaves per shoot of peach seedlings at 94 days after planting as affected by vermicompost addition to the growing substrate and AMF inoculation at transplanting time. Different letters in the factor indicate statistical differences by Tukey ($\alpha = 0.05$). $n = 34$.

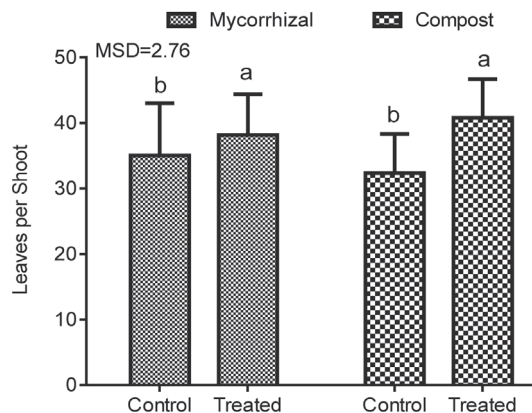


Figure 4. Root colonization by AMF (*Glomus* spp.) in peach seedlings as affected by vermicompost addition to the growing substrate and AMF inoculation at transplanting time. Data from plants after 180 days of transplanting. M0C0, control; M1C0, AMF-inoculated in substrate without vermicompost; M0C1, non-AMF-inoculated in substrate with vermicompost; and M1C1 AMF-inoculated in substrate with vermicompost. Different letters indicate statistical differences by Tukey ($\alpha = 0.05$). $n = 7$.

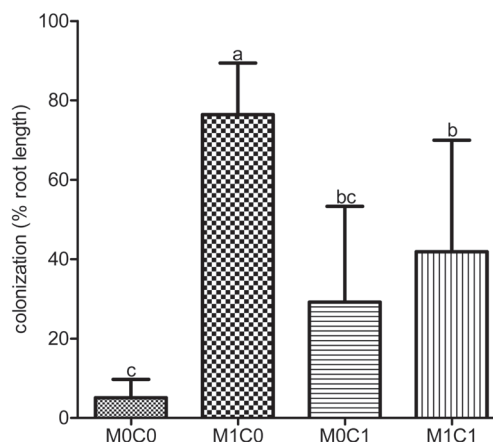


Table 3. Leaf element (N, P, and K) concentrations (dry weight basis) in peach seedlings grown in media either with or without vermicompost and either inoculated or not inoculated with the *Glomus* AM fungi. Data from plants after 180 days from germination/inoculation (n = 7).

	Mineral leaf concentration (%)		
	N ²	P	K
With vermicompost			
Non inoculated	2.19	0.26	3.08
AMF inoculated	2.11	0.24	3.00
Without vermicompost			
Non inoculated	2.17	0.16	2.33
AMF inoculated	1.68	0.32	2.28
Analysis of variance			
Mycorrhizae	**	*	Ns
Vermicompost	**	ns	**
Mycorrhizae x vermicompost	**	**	Ns

² N was determined by titration with H₂SO₄ (0.047N) after digestion with H₂SO₄ and distillation in NaOH (40%); P was measured with a spectrophotometer after wet digestion; and K was measured by flame spectrometry.

ns, non-significant; *, significant (p < 0.05); and **, high significance (p < 0.01) by Tukey test.

Previous research has shown a decrease in leaf N concentration in peach in response to mycorrhizal inoculation (La Rue *et al.*, 1975) and there is evidence that AMF inoculation does not promote N acquisition in some herbaceous perennial plants and can even suppress the plant acquisition for this mineral (Reynolds *et al.*, 2005). In this experiment, AMF inoculation showed significant effect on shoot height but not on dry matter distribution (Figure 1 and Table 2) meaning that growth of mycorrhizal peach seedlings was supported more by cell elongation and water than by assimilates including all N forms.

When plants grown in substrate without vermicompost are compared, mycorrhizal inoculation also had significant effect on leaf P concentration as AMF inoculated plants had higher leaf P concentration than non-AMF inoculated plants (Table 3). Mycorrhizal fungi are

known as symbiotic microorganisms which improve plant P uptake. Phosphorous is a low mobile mineral in the soil and depletes rapidly near the root system and the beneficial effect of mycorrhizal fungi on P plant uptake arises from the rapid growth of the extraradical mycelium beyond its depletion zone (Kaschuk *et al.*, 2009). Regarding K, mycorrhizal inoculation did not affect its leaf concentration. Previous works have shown that mycorrhizal inoculation may not affect the leaf concentration of this mineral neither in peach (La Rue *et al.*, 1975) nor in *Citrus tangerine* seedlings at moderate temperatures (Wu and Zou, 2010). The beneficial effect of the mycorrhizal inoculation on K uptake might be found when plants are grown under certain stress factors like temperature in citrus (Wu and Zou, 2010), salinity in tomato (Hajiboland *et al.*, 2010) and drought in *Arbutus unedo* (Navarro-García *et al.*, 2011). In this experiment, mineral status and salinity of the substrate, as well as temperature in the glasshouse and irrigation schedule were not stress factors; thus, peach seedlings might have absorbed K without limits.

In this experiment, vermicompost addition to the substrate resulted in significantly higher concentrations N and K in peach leaves but did not affect P leaf concentration. Other authors mention no effect of compost on peach N, P and K leaf concentrations during the first three years of growth (Baldi *et al.*, 2006). Also, other authors found a linear increase in foliar N and P as a result of increasing the rate of composted turkey litter in the substrate for potted *Cotoneaster* and *Hemerocallis*, but foliar K was not affected (Tyler *et al.*, 1993). Compost effect on plant depends upon its quality, which results from the material and process used during composting, thus meaning that compost quality may be too variable (Raviv, 2005).

Regarding root colonization, AMF inoculated plants growing in media without vermicompost resulted in 76% of the root colonized by the fungus after six months from inoculation (Figure 4). Previous reports had mentioned that peach is heavily colonized by the AMF *Glomus* spp.,

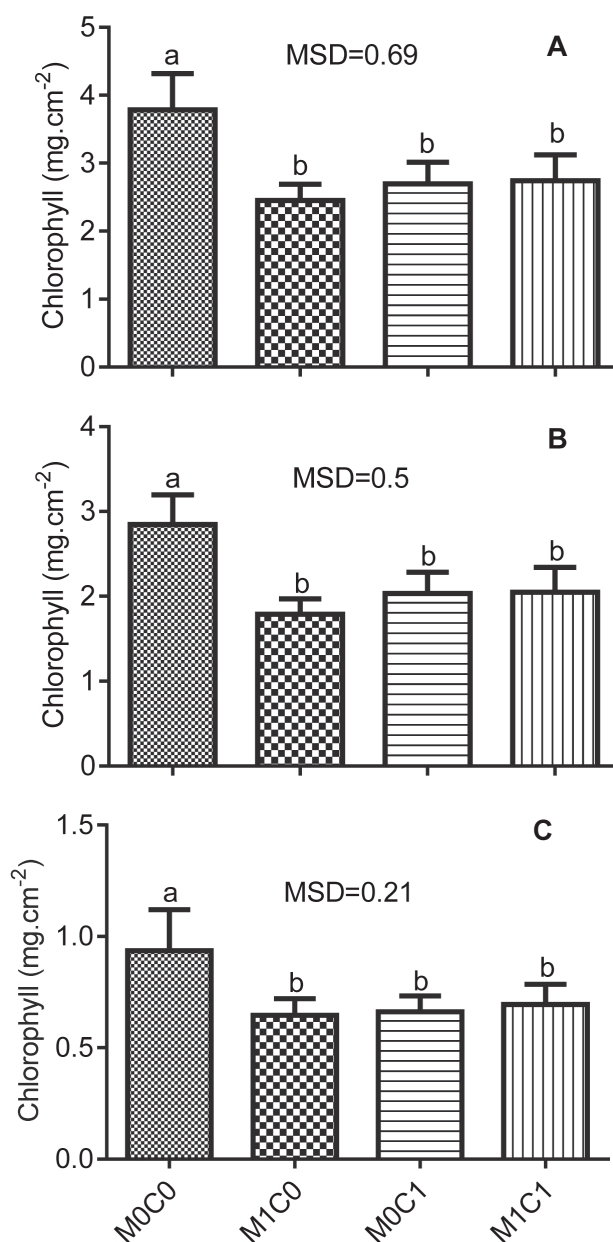
leading to root colonization from 60% to 71% depending on the peach genotype (Traquair and Berch, 1988). This high root colonization by *Glomus* spp. was also observed in seven-month-old peach plants growing in a sterilized peat-sand media (Rapparini *et al.*, 1994). Therefore, such high root colonization by the AMF was expected in this experiment with autoclaved substrates without vermicompost; however, when we compare inoculated plants growing in substrate with vermicompost to those inoculated plants growing in substrate without vermicompost, it can be observed that adding vermicompost to the substrate resulted in statistically lower root AMF colonization. Regarding this, some authors had found that high organic matter levels in substrates are less conducive to AMF colonization in peach (Morrison *et al.*, 1993; Estaún *et al.*, 1999). Non-inoculated plants with vermicompost also resulted in 29.2% of root colonized by the AM fungus which was expected as vermicompost was not autoclaved.

Control plants resulted in higher concentrations of both chlorophylls *a* and *b* and, consequently, with higher concentration of total chlorophyll (Figure 5).

These plants were always the shorter plants throughout the experiment (Figure 1) and had one of the higher N leaf contents (Table 2). Nitrogen is necessary for chlorophyll synthesis (Kaschuk *et al.*, 2009) and the higher nitrogen in those control plants could lead to higher chlorophyll synthesis; additionally, the more intensive growth in AMF inoculated plants and all those plants grown in substrate with vermicompost could result in a dilution effect of this pigment and less concentration per foliar area registered in the tallest plants.

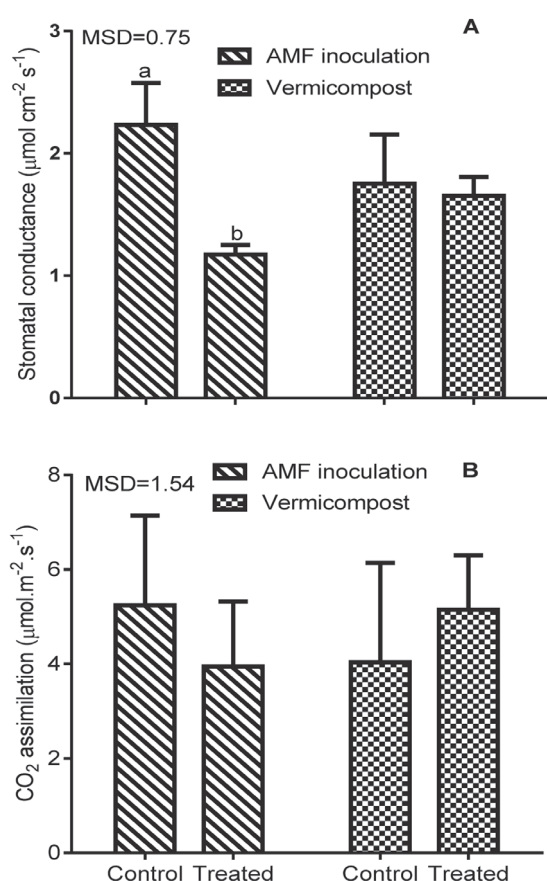
Intensive growth also results in more demand for carbohydrates as the sink strength increases and this can result in photosynthesis increases (Kaschuk *et al.*, 2009); however, in this experiment, the lesser concentrations of chlorophylls in those plants with more intensive growth compared with the control plants could eclipse this effect (Figure 6B).

Figure 5. Total chlorophyll (A), chlorophyll *a* (B), and chlorophyll *b* (C) contents in peach seedlings leaves as affected by vermicompost addition to the growing substrate and AMF inoculation at transplanting time. Data from plants after 108 days of transplanting. M0C0, control; M1C0, AMF-inoculated in substrate without vermicompost; M0C1, non-AMF-inoculated in substrate with vermicompost; and M1C1 AMF-inoculated in substrate with vermicompost. Different letters in the chlorophyll type indicate statistical differences by Tukey ($\alpha = 0.05$). $n = 5$.



Contrary to other reports which have found a positive effect of the AMF *Glomus* on stomatal conductance in plants like tomato (Hajiboland *et al.*, 2010) and chili (Manjarrez-Martínez *et al.*, 1999), in this experiment, AMF inoculated peach plants showed a lower stomatal conductance when compared with non-AMF inoculated ones (Figure 6A). Broadley *et al.* (2001) found that leaf limited N is related to lower stomatal conductance in lettuce. These authors explained this relation on the role that N may have in leaf cell osmosis as N may act, in NO_3^- form, as an osmolite or as a signaling molecule. Comparing mycorrhizal to non-mycorrhizal plants in this experiment, leaf N concentrations were statistically lower in mycorrhizal plants and these plants also had less chlorophyll and lower stomatal conductance.

Figure 6. Stomatal conductance (A) and photosynthesis rate (B) in peach seedlings as affected by vermicompost addition to the growing substrate and AMF inoculation at transplanting time. Data from plants after 108 days from transplanting. Different letters indicate statistical differences by Tukey test ($\alpha = 0.05$; $n = 5$).



Conclusions

Both, AMF inoculation at transplanting time and vermicompost addition to the growing substrate increased shoot height in peach seedlings grown under a glasshouse, but vermicompost effects were more evident than those from AMF inoculation. The number of leaves per shoot was also increased by both AMF inoculation and vermicompost; however, these treatments had no effects on plant photosynthesis. Nevertheless, vermicompost-grown plants had higher dry weights for shoot and root which was not observed in AMF-inoculated plants.


Vermicompost in the growing substrate negatively affected root colonization by the AM fungi and increased N and K leaf concentration in those inoculated plants; however, vermicompost could not increase the concentration of N and P in the absence of mycorrhizal fungi. Vermicompost also eclipsed the benefit from mycorrhizal fungi on P uptake.

In order to stimulate peach seedling growth in the nursery after sterilizing growing materials, vermicompost addition to the substrate appears more recommended than AMF inoculation; however, because of the high interactions between AMF-inoculation and vermicompost addition to the growing substrate on some important growth and physiological variables, it is highly recommended to study the causes of these interactions in genotypes different from the one used in this experiment.

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